November 24, 1952

Dear Norton:

This is to answer yours of the 17th and 21st. I'm sorry not to have been able to get in touch with you: perhaps I would have been able to interpret your letters more accurately if I could get in better touch with what you have been doing. The paternity of SW-534 seems now pretty well established as Edwards' 157, except that I could not recover Salmonella from the tube you sent of #6, but only some gram-positive contaminant. I have written to Edwards for another sub. I don't think any detailed exposition of this vexing paternity question will ever be needed, if I can exclude #6, as should be possible. #157 was apparently isolated by Cherry 10 years ago as a second phase of a java "b:-" strain. I am trying to get the original. Its genetic behavior seems to be best explained by assuming that it derives its 1,2 character as an allele at A_1, the "specific phase" locus. For example, in 157--x abony^2 (as well as the reciprocal), one gets 12:enx (sic!), rather than the b:12 that one finds from typhimurium ^{2} -x abony, and comparable to the i:enx of tymur^1 0x abony.

So far, there does not seem to be any influence of the phase of the recipient cells on the outcome of any transductions. For example, as mentioned previously, abony^1 -x typhimurium ^{1} or 2 gives b:1,2. I would imagine that the activity of the alleles at A_1 and A_2 are mutually exclusive, and that the confused cell that becomes $A_1^* A_2^*$ by transduction must make some choice, only one outcome being immediately selected by the serum agar. One can drag the cytoplasm into the story by assuming that it imposes the state on the locus in such a way that the latter is carried over into the new cell, and then acts in turn on the cytoplasm. So far, I can find no need to invoke states other than local. Since the recipient cells are almost always contaminated with alternative phases, the work will have to be done more precisely to see whether the phase of the recipient has any effect on the efficiency of transductions to the two phases. Most of the experiments so far have been defective by reason of too dense inoculations, which overgrow and inhibit most of the transductions before the latter can swim out. With much lighter inocula, the results seem much better, and more susceptible to quantitative study. In fact, some of the buds from i -x b:- on b-agar appear to have flares, which just possibly might be b-segregants from an intermediate (b)i state.

I agree with your remarks on the scope of the ms. that Bruce is now brooding over. There would be no harm in casual mention of any point that is immediately pertinent, but there will be no end to it if we practise free association in organizing it.

Concerning the relationship of phage to transduction, the PLT22 / SW-666 (543 Gal -) system may be very useful indeed. The host adaptation of PLT22 to SW666 is apparently a mutation which persists when the "PLT22B" is grown again on LT2. It is not difficult to make SW666 lysogenic for 22B, without decreasing its transinduceability (likely increasing it 2-3X). Anyhow, there seems to be a definite correlation between Gal+ transduction and lysogenization. Papillae from 22B(543) -x 666 were picked, and the purified Gal+ compared with the contaminating Gal-, and with interpapillary Gal-, for lysogenicity. The closest

comparison is between the Gal+ and the Gal- from the same streakings. There did not seem to be any correlation (the main point of the three point comparison) as would indicate a common descent of individual + and -. The results give the following table:

	Lp^+	Lp^-
Gal+	18	13
Gal-	5	15

which gives a X^2, p between .05 and .01.

Any bias seems, however, to be against a difference, as the Gal^- may well have been reinfected by the Gal^+ in a few cases. If one adds the interpapillary Gal-, the difference is very marked, as the incidence of Lp^+ there is only about 10%. A better control is still needed, however, a comparison of added, marked Gal+ Lp^s recovered from the same plates as the transductions. The results already point to a distinct correlation of the induction of lysogenicity with transduction. Whatever this is due to, and especially if it is a matter of insufficient phage to saturate the plates, it does point again to FA-phage. With most other systems, the efficiency of lysogenization is too high to do this experiment. *See PS.

The points that I am working on now are a) the determination of phases, as mentioned; b) the curious genetics of #157; c) the PLT22B story as above and d) the determination of b/i ratio in the linked transduction. The two possibilities that are still unsettled for the latter are that: a) fragments from an SW-543 derivative are always larger than from LT-2, or b) the selection for linkage in the first transduction (tymur -x 543)to give i has resulted in a tighter association of the linked loci in subsequent transductions, You will recall that typhimurium -x 543 gives mostly b, while the(tymur -x 543)i -x 543 gives mostly i. I have some two-step transductions, tymur --x (tymur -x 543)b whose FA's behavior should settle this question. Spicer has some Group E phages now, but they adsorb poorly and apparently do not transduce. More are being tried.

I am glad you have cleared up the SW541-565 story; are you sure of it? I was fairly sure I had purified 541 before setting up the mutant isolation; perhaps it is unstable. Also, some transductions LT2 -x 565 are full +/-? I am not sure I know what lytic variant you are talking about. How do you define a lytic variant? We thought once that lambda was lytic for 123, but it turns out to have been "host-modified". Have you answered the question of your 5th paragraph, 11/17 ?

Your respreading experiment is interesting. Did you not get some similar results last Spring? How about a more detailed account of this one: did you study Gal+ and S^r simultaneously, or only the latter? The delay in SM action is expected, judging from killing curves; Newcombe's failure to find evidence of phenomic lag in spontaneous mutation, and the behavior of segregants from S^r/S^s in E. coli. You can hope that phenomic lag does not influence the transinduction counts. It might show up as an unexpectedly rapid increase in S^r when they do come through. It would be interesting to look for this using SM under optimum conditions and concentrations for immediate effects. Do you believe there are any phage-resistance markers in Salmonella excepting lysogenicity and S-R mechanisms? I don't quite see how replica plating can help very much use. You can only transfer 1-10% at a time. Just what sort of application did you have in mind?

The big question will be to prove that the delay in increase of transinductions is due to segregation rather than an irregular inhibition of growth. If something of the order of 1/100 of your cells are transinduced for some character, and these are inhibited, it would be difficult to determine whether just these cells are, for some reason, differentially delayed.(1/100 is guessed as 10^{-6} [eff. of transd/trait] x 10^4 number of traits).

P.S. An interesting sideline: A few of the SW-666 x- 543, Gal+, are apparently unstably lysogenic. The colonies have a very rough appearance, rarely throw smooth-looking Lp^s. This has been seen before, but rarely so clearly. I expect that early reports (e.g. Deskowitz) on unstable R-S variation may have been based on this sort of thing. It can be predicted too that the "rough" Lp^s/s will be hemolytic. Joe Bertani has noticed something similar in Shigella. I don't think these types are likely to be diploid (they do not segregate Gal concurrently in the tests so far), but may be clues to the intermediate states preceding lysogenic stability.

Sincerely,

Joshua Lederberg